**EXHIBIT 25**



1: Anal Biochem. 2003 Nov 15;322(2):225-32.

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# Cellodextrin preparation by mixed-acid hydrolysis and chromatographic separation.

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A procedure for preparation of purified cellodextrins in gram quantities was developed for use in biochemical and microbiological studies. Cellodextrins were prepared by hydrolyzing microcrystalline cellulose (Avicel) over a period of 4 to 5.5h in the presence of a mixture of 80% (v/v) concentrated hydrochloric acid ( approximately 37 wt.%) and 20% (v/v) concentrated sulfuric acid ( approximately 98 wt.%) at room temperature (22 degrees C). Acetone precipitation, washing ion exchange, and neutralization with barium hydroxide were used to generate a solution of mixed cellodextrins substantially free of acids and salts. Yields following hydrolysis and precipitation were approximately 0.05, approximately 0.07, approximately 0.06, and approximately 0.02 g/g cellulose for cellotriose (G(3)), cellotetraose (G(4)), cellopentose (G(5)), and cellohexose (G(6)), respectively. Cellodextrins with degrees of polymerization from 3 to 11 were separated chromatographically using a 29 x 5-cm I.D. Bio-Rad AG50W-X4 column arranged in series with a 91 x 5-cm I.D. Bio-Gel P4 column. This two-column system was used to obtain cellodextrin preparations at 240 mg/day for G(3), 330 mg/day for G (4), 260 mg/day for G(5), and 130 mg/day for G(6), with purity >99% for G(3), G(4), and G(5) and >95% for G(6). The overall procedure achieves yields comparable to the highest previously reported, employs a separation system that can readily be reused for multiple runs, and avoids use of fuming HCl.

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